

METHOD FOR INCREASING THE BIOAVAILABILITY OF THE ACTIVE FORM OF
S-[2-([[[1-(2-ETHYLBUTYL)CYCLOHEXYL]CARBONYL]AMINO)PHENYL] 2-
METHYLPROPANETHIOATE

CROSS-REFERENCE TO RELATED APPLICATIONS

[0001] This patent application claims the benefit of U.S. Provisional Patent Application Nos. 60/455,293, filed March 17, 2003; 60/460,521, filed April 4, 2003; and 60/477,202, filed June 10, 2003.

FIELD OF THE INVENTION

[0002] This invention is directed to methods for the treatment or prophylaxis of cardiovascular disorders.

BACKGROUND OF THE INVENTION

[0002] Hyperlipidemic conditions associated with elevated concentrations of total cholesterol and low-density lipoprotein (LDL) cholesterol are major risk factors for coronary heart disease, and atherosclerosis in particular. Additionally, numerous studies have demonstrated that a low plasma concentration of high-density lipoprotein (HDL) cholesterol is a powerful risk factor for the development of atherosclerosis.

[0003] Cholesteryl ester transfer protein (CETP) is a plasma protein that facilitates the movement of cholesteryl esters and triglycerides between various lipoproteins in the blood. The movement of cholesteryl ester from HDL to LDL by CETP has the effect of lowering HDL cholesterol and increasing LDL cholesterol. Inhibition of CETP activity has been shown to effectively modify plasmid HDL/LDL ratios by elevating plasma HDL cholesterol and lowering plasma LDL cholesterol.

[0004] *S*-[2-([[(1-(2-ethylbutyl)cyclohexyl]carbonyl]amino)phenyl] 2-methylpropanethioate has been shown to be an inhibitor of CETP activity in humans (de Groot et al., *Circulation*, 105, 2159-2165 (2002)) and rabbits (Shinkai et al., *J. Med. Chem.*, 43, 3566-3572 (2000); Kobayashi et al., *Atherosclerosis*, 162, 131-135 (2002); and Okamoto et al., *Nature*, 406(13), 203-207 (2000)). *S*-[2-([[(1-(2-ethylbutyl)cyclohexyl]carbonyl]amino)phenyl] 2-methylpropanethioate has been shown to increase plasma HDL cholesterol in humans (de Groot et al., *supra*) and in rabbits (Shinkai et al., *supra*; Kobayashi et al., *supra*; Okamoto et al., *supra*). Moreover, *S*-[2-([[(1-(2-ethylbutyl)cyclohexyl]carbonyl]amino)phenyl] 2-methylpropanethioate has been shown to decrease LDL cholesterol in humans (de Groot et al., *supra*) and rabbits (Okamoto et al., *supra*). Additionally, *S*-[2-([[(1-(2-ethylbutyl)cyclohexyl]carbonyl]amino)phenyl] 2-methylpropanethioate inhibits the progression of atherosclerosis in rabbits (Okamoto et al.,

supra). *S*-[2-([[1-(2-ethylbutyl)cyclohexyl]carbonyl]amino)phenyl] 2-methylpropanethioate, as well as methods of making and using the compound, are described in U.S. Patent 6,426,365.

[0005] It would be desirable to increase the bioavailability of the active form of *S*-[2-([[1-(2-ethylbutyl)cyclohexyl]carbonyl]amino)phenyl] 2-methylpropanethioate to increase the extent of the therapeutic effect on the user. There remains a need for a method of doing so, for example, a method of treating cardiovascular disorders using *S*-[2-([[1-(2-ethylbutyl)cyclohexyl]carbonyl]amino)phenyl] 2-methylpropanethioate that has increased bioavailability of its active form and, thus, improved efficacy following oral administration to patients. This invention provides such a method, as well as a kit comprising a pharmaceutical composition comprising a therapeutically effective amount of *S*-[2-([[1-(2-ethylbutyl)cyclohexyl]carbonyl]amino)phenyl] 2-methylpropanethioate and a pharmaceutically acceptable carrier, prescribing information, and a container, wherein the prescribing information includes advice to a patient regarding administration of the drug to improve bioavailability. These and other advantages of the invention, as well as additional inventive features, will be apparent from the description of the invention provided herein.

BRIEF SUMMARY OF THE INVENTION

[0006] The invention provides a method of increasing the bioavailability of the active form of *S*-[2-([[1-(2-ethylbutyl)cyclohexyl]carbonyl]amino)phenyl] 2-methylpropanethioate in a patient receiving *S*-[2-([[1-(2-ethylbutyl)cyclohexyl]carbonyl]amino)phenyl] 2-methylpropanethioate therapy wherein *S*-[2-([[1-(2-ethylbutyl)cyclohexyl]carbonyl]amino)phenyl] 2-methylpropanethioate is contained in a pharmaceutical composition, which method comprises orally administering a therapeutically effective amount of the drug to the patient with food. The increased bioavailability of the active form can be evidenced by an increase in the maximum plasma concentration of the active form.

[0007] The invention also provides a method of increasing the extent of absorption of that active form of *S*-[2-([[1-(2-ethylbutyl)cyclohexyl]carbonyl]amino)phenyl] 2-methylpropanethioate as measured by the concentration of the active form attained in the blood stream over time of a patient receiving the drug in an oral dosage form. This method comprises orally administering a therapeutically effective amount of *S*-[2-([[1-(2-ethylbutyl)cyclohexyl]carbonyl]amino)phenyl] 2-methylpropanethioate to the patient with food.

[0008] The invention further provides a method for decreasing the activity of cholesteryl ester transfer protein (CETP) in a patient. The method comprises orally administering a therapeutically effective amount of *S*-[2-([[1-(2-

ethylbutyl)cyclohexyl]carbonyl]amino)phenyl] 2-methylpropanethioate to the patient with food.

[0009] The invention additionally provides a method for the treatment or prophylaxis of a cardiovascular disorder in a patient. The method comprises orally administering *S*-[2-([1-(2-ethylbutyl)cyclohexyl]carbonyl]amino)phenyl] 2-methylpropanethioate to the patient with food.

[0010] Moreover, the invention provides a kit comprising a pharmaceutical composition comprising a therapeutically effective amount of *S*-[2-([1-(2-ethylbutyl)cyclohexyl]carbonyl]amino)phenyl] 2-methylpropanethioate and a pharmaceutically acceptable carrier, prescribing information, and a container. The prescribing information includes advice to a patient regarding the administration of *S*-[2-([1-(2-ethylbutyl)cyclohexyl]carbonyl]amino)phenyl] 2-methylpropanethioate with food.

BRIEF DESCRIPTION OF THE DRAWINGS

[0011] Figure 1 is a linear plot of the geometric mean plasma concentrations ($\mu\text{g/mL}$) of the active form of *S*-[2-([1-(2-ethylbutyl)cyclohexyl]carbonyl]amino)phenyl] 2-methylpropanethioate over 36 hours in Caucasian male patients, who were orally administered 900 mg of *S*-[2-([1-(2-ethylbutyl)cyclohexyl]carbonyl]amino)phenyl] 2-methylpropanethioate with food or without food.

[0012] Figure 2 is a semi-logarithmic plot of the geometric mean plasma concentrations ($\mu\text{g/mL}$) of the active form of *S*-[2-([1-(2-ethylbutyl)cyclohexyl]carbonyl]amino)phenyl] 2-methylpropanethioate over 36 hours in Caucasian male patients, who were orally administered 900 mg of *S*-[2-([1-(2-ethylbutyl)cyclohexyl]carbonyl]amino)phenyl] 2-methylpropanethioate with food or without food.

[0013] Figure 3 is a plot of the mean changes from baseline (pre-dose) in CETP activity over 24 hours in Caucasian male patients, who were orally administered 900 mg of *S*-[2-([1-(2-ethylbutyl)cyclohexyl]carbonyl]amino)phenyl] 2-methylpropanethioate with food or without food.

[0014] Figure 4 is a plot of mean CETP activities and mean plasma concentrations of the active form of *S*-[2-([1-(2-ethylbutyl)cyclohexyl]carbonyl]amino)phenyl] 2-methylpropanethioate over 24 hours in Caucasian male patients following the oral administration of 900 mg of *S*-[2-([1-(2-ethylbutyl)cyclohexyl]carbonyl]amino)phenyl] 2-methylpropanethioate with food.

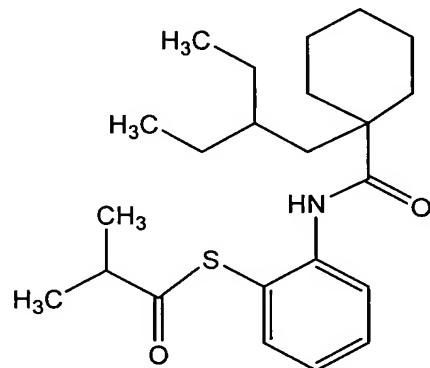
[0015] Figure 5 is a plot of mean CETP activities and mean plasma concentrations of the active form of *S*-[2-([1-(2-ethylbutyl)cyclohexyl]carbonyl]amino)phenyl] 2-methylpropanethioate over 24 hours in Caucasian male patients following the oral

administration of 900 mg of *S*-[2-(([1-(2-ethylbutyl)cyclohexyl]carbonyl)amino)phenyl] 2-methylpropanethioate without food.

DETAILED DESCRIPTION OF THE INVENTION

[0016] The invention is directed to methods for improving the effectiveness of the administration of *S*-[2-(([1-(2-ethylbutyl)cyclohexyl]carbonyl)amino)phenyl] 2-methylpropanethioate (herein referred to as Compound I) and/or for the treatment or prophylaxis of certain cardiovascular disorders. These disorders include, but are not limited to, cardiovascular disease, coronary heart disease, coronary artery disease, hypoalphalipoproteinemia (low levels of HDL cholesterol), hypercholesterolemia, and atherosclerosis. Additional disorders that may be treated or prevented by the inventive methods include, but are not limited to, hyperlipidemia, hypertension, hypertriglyceridemia, and hyperlipidoproteinemia.

[0017] Compound I has the following structural formula:



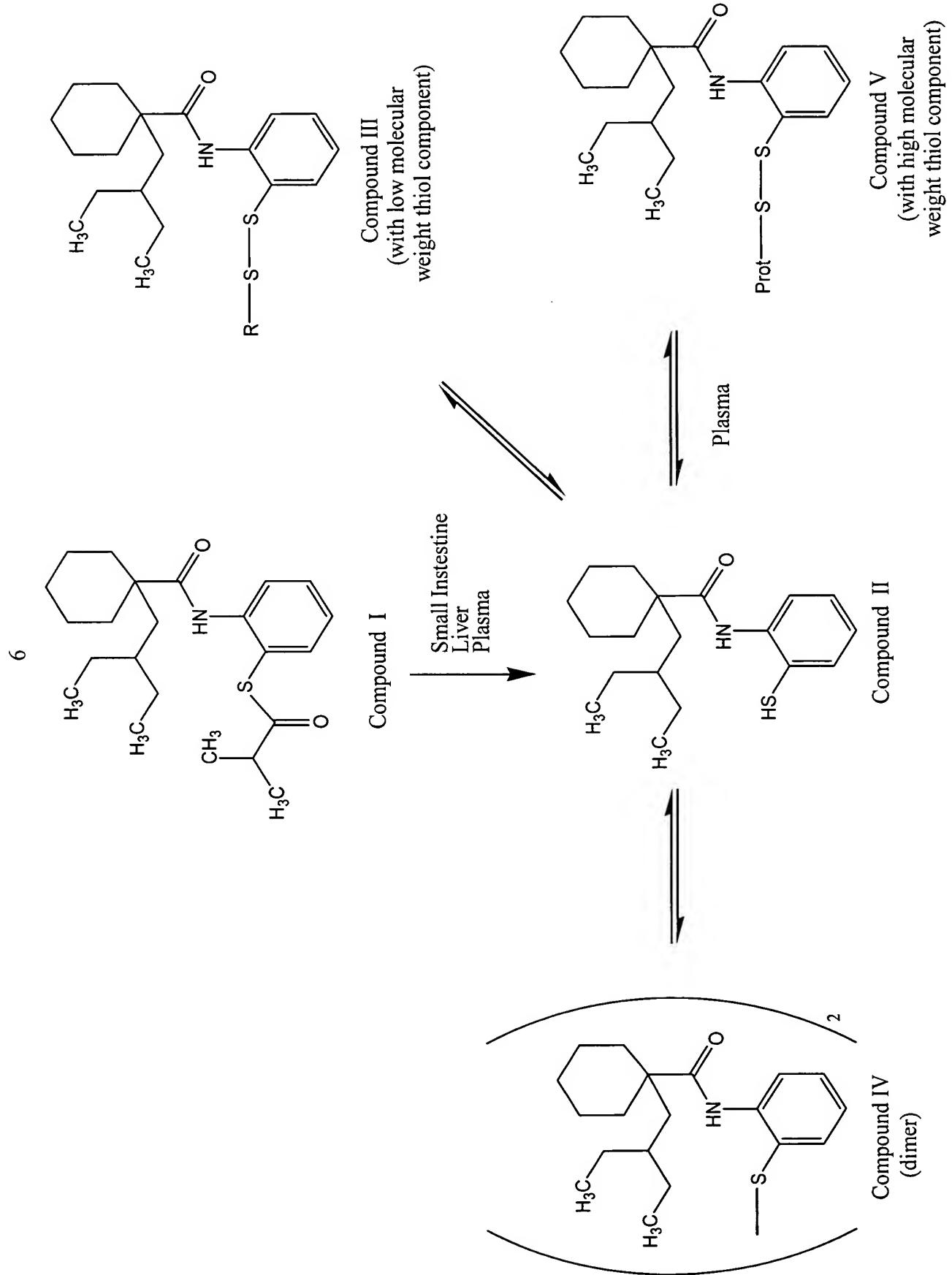
Pharmacokinetic studies have not previously been conducted to evaluate the effect of food on the pharmacokinetics of the active form of Compound I. In general, food has a variable effect on the bioavailability of an active agent. Drug-food interactions may result in reduced, delayed, or increased systemic drug availability. See, e.g., Welling, *Clin. Pharmacokinet.*, 9(5), 404-34 (1984).

[0018] It has been discovered that Compound I can be administered to patients in a regimen that increases the therapeutic effectiveness of Compound I to such patients. Advantageously, when orally administered with food, Compound I exhibits increased bioavailability of the active form of Compound I in patients.

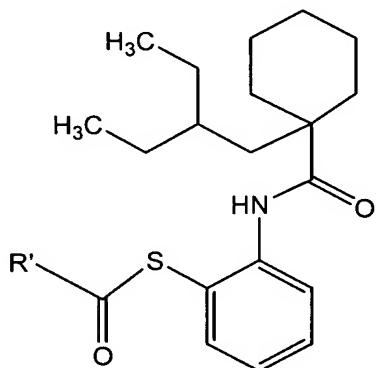
[0019] Accordingly, the invention provides a method of increasing the bioavailability of the active form of Compound I in a patient comprising administering to the patient a therapeutically effective amount of Compound I with food.

[0020] As used herein, the term "bioavailability" generally means the rate and extent to which the active ingredient, or active form thereof, is absorbed from a drug product and becomes available at the site of action. See U.S. Code of Federal Regulations, Title 21, Part 320.1 (2001 ed.). For oral dosage forms, bioavailability relates to the processes by which the active ingredient is released from the oral dosage form, e.g., a tablet, converted to the active form (if the active ingredient is not already the active form), and moved to the site of action, e.g., absorbed into the systemic circulation.

[0021] While not wishing to be bound by any particular theory, it is hypothesized that within the body of a patient, Compound I is hydrolyzed in plasma, the liver, and/or the small intestine to form *S*-[2([[1-(2-ethylbutyl)cyclohexyl]carbonyl]amino)phenyl] thiol (herein referred to as Compound II). It is known that low molecular weight thiol components (i.e., R-SH), such as cysteine and glutathione, and high molecular weight thiol components (i.e., Prot-SH), such as peptides and proteins (e.g., enzymes and cell membranes), exist in the body as mixed disulfides containing an oxidized disulfide bond (S-S bond) between or within the molecule (see, e.g., Shimade et al., *J. Chromatogr. B*, 659, 227 (1994)). Therefore, it is hypothesized that within the body of a patient, Compound II is conjugated with low or high molecular weight thiols to yield mixed disulfides or to yield dimers of Compound II. Since these forms are in an oxidation-reduction equilibrium with each other via Compound II, all of these forms, as well as Compound II, are collectively, but not exclusively, considered and referred to hereafter as the active form of Compound I. The following scheme depicts the above-described hypothesis.



[0022] While the administration of Compound I is a particularly preferred embodiment of the invention, the invention also contemplates the administration of other compounds that will yield the active form of Compound I, i.e., other prodrugs of the active form of Compound I. Such prodrugs, for example, can be compounds that have different mercapto-protecting groups, but that still result in the formation of the active form of Compound I (e.g., Compound II) in the body of a patient (i.e., *in vivo*). The term “mercapto-protecting groups” refers to commonly used mercapto-protecting groups (e.g., as described in Wolman, *The Chemistry of the Thiol Group*, D. Patai, Ed., Wiley-Interscience, New York, 1974). Any organic residues that can be dissociated *in vivo* may be used without particular restriction. Examples of particularly suitable mercapto-protecting groups are described in U.S. Patent 6,426,365. The invention further contemplates the administration of Compound I' (wherein R' signifies an organic residue other than an isopropyl group) so as to yield the active form of Compound I.



Compound I'

In addition, Compounds III, IV, and V (wherein R signifies an organic residue and Prot signifies a peptide or protein), which are believed to be in equilibrium with Compound II *in vivo*, similarly can be directly administered to the patient.

[0023] As used herein, the term “with food” is defined to mean, in general, the condition of having consumed food during the period between from about 1 hour prior to the administration of Compound I to about 2 hours after the administration of Compound I. Preferably, the food is a solid food with sufficient bulk and fat content that it is not rapidly dissolved and absorbed in the stomach. More preferably, the food is a meal, such as breakfast, lunch, or dinner.

[0024] Advantageously, Compound I is administered any time of day with food. The food can be consumed at any time during the period between from about 1 hour prior to the administration of Compound I to about 2 hours after the administration of Compound I. For example, the food can be consumed within the time period of about 1 hour, about 45

minutes, about 30 minutes, about 15 minutes, about 10 minutes, or about 5 minutes prior to the administration of Compound I. Similarly, the food can be consumed within the time period of about 5 minutes, about 10 minutes, about 15 minutes, about 30 minutes, about 45 minutes, about 1 hour, about 1.25 hours, about 1.5 hours, about 1.75 hours, or about 2 hours after the administration of Compound I. More preferably, the administration of Compound I to the patient is immediately after the consumption of food (e.g. within about 1 minute after food consumption) up to about 1 hour after food consumption. Ideally, Compound I is administered at substantially the same time as the consumption of the food.

[0025] The terms “without food” or “fasted” are defined to mean the condition of not having consumed food within the time period of about 1 hour prior to the administration of Compound I to about 2 hours after the administration of Compound I.

[0026] The term “patient” refers to a human patient.

[0027] The methods of the invention are directed to the administration of a therapeutically acceptable amount of Compound I or a prodrug of the active form of Compound I, e.g., a prodrug of Compound II. In general, a suitable dose of a therapeutically effective amount of Compound I for administration to a patient will be between approximately 100 mg to about 1800 mg per day. A suitable dose is preferably about 300 mg to about 900 mg per day. A preferred dose is about 600 mg per day.

[0028] If desired, the effective daily dose of Compound I may be administered as two, three, four, five, six, or more sub-doses administered separately at appropriate intervals throughout the day, optionally, in unit dosage forms. Each such sub-dose contains a therapeutically effective amount of Compound I. In accordance with the inventive method, Compound I may be administered with food at multiple times per day or, alternatively, once per day. When administered at multiple times throughout the day, each individual dose contains a therapeutically effective amount of Compound I. In a preferred embodiment of the invention, Compound I is administered with food once per day.

[0029] As used herein, the term “unit dosage form” is defined to refer to the form in which Compound I is administered to the patient. Specifically, the unit dosage form can be, for example, a pill, capsule, or tablet. Preferably, the unit dosage form is a tablet. The typical amount of Compound I in a unit dosage form useful in the invention is about 100 mg to about 1800 mg, preferably about 100 mg to about 900 mg (e.g., about 100 mg to about 300 mg). In a preferred embodiment of the invention, the unit dosage form comprises about 300 mg of Compound I and is in the form of a tablet. Preferably, two or three tablets, each of which comprises about 300 mg of Compound I, are administered to the patient once per day (i.e., a total dose per day of about 600 mg or about 900 mg, respectively).

[0030] The increased bioavailability of the active form of Compound I to a patient receiving Compound I therapy can be evidenced in any suitable manner. Desirably, the oral

administration of Compound I with food results in an increased bioavailability of the active form of Compound I as evidenced by an increase in the maximum plasma concentration of the active form of Compound I as compared to the administration of Compound I without food.

[0031] Compound I desirably is provided to a patient in a container associated with prescribing information that advises the patient to orally administer Compound I with food and desirably also explains that doing so will increase the bioavailability of the active form of Compound I. Compound I preferably is provided to a patient in a container associated with prescribing information that advises the patient that the administration of the dose of Compound I in a pharmaceutical composition with food results in an increase in the extent of absorption of the active form of Compound I as reflected by an increase in the maximum plasma concentration of the active form of Compound I as compared to the administration of the drug under fasted conditions.

[0032] The invention also provides a method of increasing the extent of absorption of the active form of Compound I as measured by the active form concentration attained in the blood stream over time in a patient in need of a therapeutic effect thereof. This method comprises orally administering to a patient a therapeutically effective amount of Compound I in a pharmaceutical composition with food. The active form concentration in the blood stream is measured as the plasma concentration ($\mu\text{g/mL}$) of the active form of Compound I. Pharmacokinetic parameters involved in determining the plasma concentration include the maximum observed plasma concentration (C_{\max}), area under the plasma concentration time curve (AUC) from time zero up to the last quantifiable concentration ($AUC_{0-\infty}$), and AUC from time zero to infinity ($AUC_{0-\infty}$). Administering Compound I to a patient with food increases the bioavailability of the active form as measured by increased values of one or more (and desirably all) of the aforesaid pharmacokinetic parameters, when compared to administration of the drug under fasted conditions.

[0033] The invention further provides a method for decreasing the activity of cholesteryl ester transfer protein (CETP) in a patient, which comprises orally administering a therapeutically effective amount of Compound I to the patient with food. The decrease in CETP activity is greater following administration of Compound I with food versus under fasted conditions.

[0034] Additionally, the invention provides a method for the treatment or prophylaxis of a cardiovascular disorder in a patient comprising administering Compound I to the patient, which comprises orally administering a therapeutically effective amount of Compound I to the patient with food. These disorders include, but are not limited to, cardiovascular disease, coronary heart disease, coronary artery disease, hypoalphalipoproteinemia (low levels of HDL cholesterol), hypercholesterolemia, and atherosclerosis. Additional disorders

which may be treated or prevented by the inventive method include, but are not limited to, hyperlipidemia, hypertension, hypertriglyceridemia, and hyperlipidoproteinemia.

[0035] Compound I may be administered for therapy to a patient in any conventional manner. While it is possible for Compound I to be administered as the raw chemical, it is preferably administered as a pharmaceutical composition. Such pharmaceutical compositions comprise Compound I with one or more pharmaceutically acceptable carriers or excipients and optionally other therapeutic agents and/or components. The carriers or excipients must be acceptable in the sense of being compatible with the other ingredients and not deleterious to the recipient thereof. Examples of carriers or excipients for oral administration include cornstarch, lactose, magnesium stearate, talc, microcrystalline cellulose, stearic acid, povidone, crospovidone, dibasic calcium phosphate, sodium starch glycolate, hydroxypropyl cellulose (e.g., low substituted hydroxypropyl cellulose), hydroxypropylmethyl cellulose (e.g., hydroxypropylmethyl cellulose 2910), and sodium lauryl sulfate.

[0036] The pharmaceutical compositions can be prepared by any suitable method, such as those methods well known in the art of pharmacy, for example, methods such as those described in Gennaro et al., Remington's Pharmaceutical Sciences (18th ed., Mack Publishing Co., 1990), especially Part 8: Pharmaceutical Preparations and their Manufacture. Such methods include the step of bringing into association Compound I with the carrier or excipient and optionally one or more accessory ingredients. Such accessory ingredients include those conventional in the art, such as, fillers, binders, diluents, disintegrants, lubricants, colorants, flavoring agents, and wetting agents.

[0037] The pharmaceutical compositions can provide controlled, slow release, or sustained release of the Compound I over a predetermined period of time. The controlled, slow release, or sustained release of the therapeutic compound can provide for a concentration of the active form of Compound I to be maintained in the bloodstream of the patient for a longer period of time than with conventional formulations. Such pharmaceutical compositions include coated tablets, pellets, and capsules, as well as dispersions of the therapeutic compound in a medium that is insoluble in physiologic fluids or where the release of the therapeutic compound follows degradation of the pharmaceutical composition due to mechanical, chemical, or enzymatic activity.

[0038] The pharmaceutical composition in the context of the invention can be, for example, in the form of a pill, capsule, or tablet, each containing a predetermined amount of Compound I and preferably coated for ease of swallowing, in the form of a powder or granules, or in the form of a solution or suspension. Preferably, the pharmaceutical composition is in the form of a tablet comprising Compound I (or a prodrug of the active form of Compound I) and the components of the tablet utilized and described in the

Examples herein. For oral administration, fine powders or granules may contain diluting, dispersing, and/or surface active agents and may be present, for example, in water or in a syrup, in capsules or sachets in the dry state, or in a nonaqueous solution or suspension wherein suspending agents may be included, or in tablets wherein binders and lubricants may be included. Components such as sweeteners, flavoring agents, preservatives (e.g., antimicrobial preservatives), suspending agents, thickening agents, and/or emulsifying agents also may be present in the pharmaceutical composition. When administered in the form of a liquid solution or suspension, the formulation can contain Compound I and purified water. Optional components in the liquid solution or suspension include suitable sweeteners, flavoring agents, preservatives (e.g., antimicrobial preservatives), buffering agents, solvents, and mixtures thereof. A component of the formulation may serve more than one function. For example, a suitable buffering agent also may act as a flavoring agent as well as a sweetener.

[0039] Suitable sweeteners include, for example, saccharin sodium, sucrose, and mannitol. A mixture of two or more sweeteners optionally may be used. The sweetener or mixtures thereof are typically present in an amount of from about 0.001% to about 70% by weight of the total composition. Suitable flavoring agents may be present in the pharmaceutical composition to provide a cherry flavor, cotton candy flavor, or other suitable flavor to make the pharmaceutical composition easier for a patient to ingest. The flavoring agent or mixtures thereof are typically present in an amount of about 0.0001% to about 5% by weight of the total composition.

[0040] Suitable preservatives include, for example, methylparaben, propylparaben, sodium benzoate, and benzalkonium chloride. A mixture of two or more preservatives optionally may be used. The preservative or mixtures thereof are typically present in an amount of about 0.0001% to about 2% by weight of the total composition.

[0041] Suitable buffering agents include, for example, citric acid, sodium citrate, phosphoric acid, potassium phosphate, and various other acids and salts. A mixture of two or more buffering agents optionally may be used. The buffering agent or mixtures thereof are typically present in an amount of about 0.001% to about 4% by weight of the total composition.

[0042] Suitable solvents for a liquid solution or suspension include, for example, sorbital, glycerin, propylene glycol, and water. A mixture of two or more solvents optionally may be used. The solvent or solvent system is typically present in an amount of about 1% to about 90% by weight of the total composition.

[0043] Oral delivery methods are often limited by chemical and physical barriers imposed by the body, such as the varying pH in the gastrointestinal tract, exposure to enzymes, and the impermeability of the gastrointestinal membranes. The oral

administration of the pharmaceutical composition may also include the co-administration of adjuvants. For example, nonionic surfactants such as polyoxyethylene oleyl ether and n-hexadecyl polyethylene ether can be administered with or incorporated into the pharmaceutical composition to artificially increase the permeability of the intestinal walls. Enzymatic inhibitors also can be administered with or incorporated into the pharmaceutical composition.

[0044] Moreover, the invention provides a kit comprising a pharmaceutical composition comprising a therapeutically effective amount of Compound I and a pharmaceutically acceptable carrier, prescribing information, and a container. The prescribing information can be prescribing information conforming to the methods of the invention and/or as otherwise discussed herein. The prescribing information preferably includes advice to a patient regarding the administration of Compound I with food, especially to improve the bioavailability of the active form of Compound I.

[0045] The following examples further illustrate the invention but, of course, should not be construed as in any way limiting its scope.

EXAMPLE 1

[0046] The effect of food on Compound I absorption in patients was identified in a study designed to compare the bioavailability of 900 mg of Compound I orally administered to Caucasian male volunteers with and without food.

[0047] For this study each of six subjects received Compound I at a dose level of 900 mg in each of two treatment periods, once with food (after a standard breakfast) and once in the fasted state. There was a minimum of 7 days between each treatment period. This interval of 7 days between treatments was considered appropriate for eliminating any within-subject carryover effects.

[0048] The subjects received 900 mg of Compound I by administration of 3 tablets of 300 mg each. The uncoated white tablets were prepared using standard tableting procedures. The tablets comprised 300 mg of Compound I, 18 mg of hydroxypropylmethyl cellulose 2910 as a binder, 18 mg of talc and 1.2 mg of magnesium stearate as lubricants, and 119.8 mg of crospovidone and 90 mg of low-substituted hydroxypropyl cellulose as disintegrants.

[0049] Treatments were administered orally with 150 mL water while standing. Subjects were not allowed to lie supine for 2 hours after dose administration, except for study procedures.

[0050] Doses were administered at similar times for each subject in each treatment period. Dosing commenced at approximately 08:30 hours. All subjects fasted from food and fluids (with the exception of water) from 22:00 hours on the day prior to dosing (Day

- 1) until breakfast on Day 1 (for subjects receiving Compound I in the fed state (i.e., with food)) or lunch-time on Day 1 (for subjects Compound I in the fasted state), and during the evening prior to the post-study visit until laboratory safety evaluations had been performed on the following day. Water could be consumed at any time during the study, with the exception of the period up to 2 hours post-dose, when no fluids were permitted.

[0051] When subjects were administered Compound I in the fed state, they received a standard breakfast about 45 minutes prior to dosing. The meal was eaten at a steady rate over a 15 minute period so that the meal was completed 30 minutes before dosing. The standard breakfast consisted of the following:

200 mL orange juice
Two packets of cereal (approximately 60 g)
Two slices of wholemeal toast
10 g low fat spread (one packet)
20 g jam (one packet)
242 mL full fat milk (approximately 250 g)

Total energy content: 711 Kcal
Total fat content: 15.72 g (19.9% of total calories)
Total protein: 20.82 g (11.7% total calories)

[0052] Blood samples for pharmacokinetic analysis were taken immediately prior to dosing and at the following times after dosing: 1, 2, 4, 6, 7, 10, 12, 24, and 36 hours post-dose.

[0053] The following pharmacokinetic parameters were calculated for the two profiles (fed and fasted conditions) and are defined as follows:

t_{max}	Time of maximum observed plasma concentration;
C_{max}	Maximum observed plasma concentration;
$t_{1/2}$	Half-life of plasma concentration of the active form of Compound I
AUC_{0-t_z}	Area under the plasma concentration-time curve (AUC) from time zero up to the last quantifiable concentration ($AUC(0-t_z)$); and
$AUC_{0-\infty}$	AUC from time zero to infinity

[0054] Pharmacokinetic parameters were log-transformed by analysis and assessed using SAS® Least Square Means derived from a three-way analysis of variance (ANOVA) fitting effects for subject, treatment, and period. Treatment comparisons were made by

calculating the difference and 95% confidence intervals (CIs) of the difference of the log SAS® Least Square Means between parameters for the respective treatments. The differences and CIs of the differences were back-transformed for reporting purposes.

[0055] The plasma concentration of the active form of Compound I was determined by the following assay. Plasma samples were isolated from patients treated with Compound I. The plasma samples were treated with sodium hydroxide (Wako Pure Chemical Industries, Ltd.) to convert active forms of Compound I in the plasma to the thiol form (i.e., Compound II). The plasma sample next was treated with dithiothreitol (DTT) (Wako Pure Chemical Industries, Ltd.) to prevent the oxidation of thiol groups (i.e., to maintain thiol groups in a reduced state). N-ethylmaleimide (NEM) (Wako Pure Chemical Industries, Ltd.) was added to stabilize the thiol form (i.e., Compound II) by, it is believed, blocking the free sulphydryl group by the derivatization to an NEM-adduct. The sample then was analyzed using High Performance Liquid Chromatography (HPLC). Finally, the results of the HPLC analysis of the plasma sample were compared to a known standard to determine the plasma concentration of the active form of Compound I. The standard of known concentration was prepared essentially as described above, with the exception that human plasma was isolated from humans who were not treated with Compound I. These “blank plasma” samples were combined with a known amount of Compound I.

[0056] The mean test results for plasma pharmacokinetic parameters, $AUC_{0-\infty}$ ($\mu\text{g h/mL}$), AUC_{0-tz} ($\mu\text{g h/mL}$), C_{max} ($\mu\text{g/mL}$), $t_{1/2}$ (h), and t_{max} (h), of the active form of Compound I are summarized in Table 1.

Table 1 – Plasma Pharmacokinetic Parameters of the Active Form of S-[2-([1-(2-ethylbutyl)cyclohexyl]carbonyl)amino]phenyl] 2-methylpropanethioate

Parameter	Treatment Protocol		Ratio (Fed:Fasted)
	Fasted	Fed	
AUC (0-t _z) *	6.21 (μg h/mL)	10.2 (19.0)	1.65
AUC (0-∞) *	7.97 (μg h/mL)	12.5 (17.4)	1.57
C _{max} *	0.423 (μg/mL)	0.955 (26.1)	2.26
t _{max} (h) ⁺	5.00 (2.00-6.00)	4.00 (2.00-6.00)	NA
t _{1/2} (h) [‡]	16.5 (14.1-22.4)	15.4 (12.6-18.6)	0.935

* = geometric mean (geometric coefficient of variation %)

⁺ = median (min-max)

NA = not applicable

‡ = harmonic mean (min-max)

[0062] The absorption of the active form of Compound I was relatively slow, with the time of maximum observed plasma concentration occurring at between 4 and 5 hours after administration of Compound I. As is apparent from Table 1, the time of maximum observed plasma concentration was similar after administration of Compound I with and without food. Additionally, the half life of the active form of Compound I was determined to be similar after administration of the drug with and without food.

[0063] Several of the pharmacokinetic parameters, however, were affected by the administration of Compound I with food. These include AUC_{0-t_z}, AUC_{0-∞}, and C_{max}, which were 65%, 57%, and 126% higher, respectively, when Compound I was administered with food as compared with the administration of Compound I in the fasted state. These increases are noticeably apparent when the geometric mean plasma concentrations of the active form of Compound I were plotted in linear form in Figure 1 and plotted in semi-logarithmic form in Figure 2.

[0064] The observed increases in pharmacokinetic parameters when Compound I is administered with food indicate an increase in the bioavailability of the active form of the drug when compared to administration of the drug under fasted conditions.

EXAMPLE 2

[0057] The effect of food on the absorption of the active form of Compound I in Caucasian male patients was identified in a study designed to compare the CETP activity following the oral administration of 900 mg of Compound I with and without food.

[0058] Administration, dosing, and sampling schedules were substantially similar to those described in Example 1.

[0059] The procedure for determining CETP activity was substantially similar to the procedures described in Tollefson et al., *Methods Enzymol.*, 129, 797-816 (1986), and Kato et al., *J. Biol. Chem.*, 264, 4082-4087 (1989).

[0060] CETP activity and changes from baseline (pre-dose) were measured, and the resulting data is summarized in Table 2 as percentage change from baseline. The mean changes from baseline (pre-dose) in CETP activity over time are set out in the plot of Figure 3.

Table 2 – Mean (S.D.) Changes from Baseline (Pre-dose) in CETP Activity

Treatment Protocol	Percent Changes from Pre-dose (standard deviation)							
	Pre-dose	1 h	2 h	4 h	6 h	8 h	24 h	Post-study (all subjects)
Fed	96 (17.0)	-1 (2.3)	-16 (13.2)	-44 (14.0)	-59 (11.1)	-58 (11.1)	-34 (7.5)	96 (17.4)
Fasted	91 (16.0)	1 (2.2)	2 (2.4)	-4 (2.2)	-10 (3.1)	-15 (4.7)	-10 (3.8)	

[0061] A clear difference in CETP activity was observed when Compound I was administered with and without food. Inhibition of CETP activity was much more marked in the fed treatment protocol as compared with the fasted treatment protocol. For example, between 4 and 24 hours post-dose, there is a significant decrease in CETP activity in the fed versus the fasted state. Such a decrease in CETP activity indicates increased bioavailability of the active form of the drug when administered with food as compared to administration of the drug without food.

[0062] The relationships between plasma concentrations of the active form of Compound I and inhibition of CETP activity for the fed and fasted states are illustrated by the plots of Figures 4 and 5, respectively. As plasma concentrations of the active form of Compound I increased, the inhibitory effect on CETP increased (i.e., CETP activity decreased).

EXAMPLE 3

[0063] In a similar study to that described in Example 2, the effect of food on the absorption of the active form of Compound I in Japanese male patients was identified in a study designed to compare the relative CETP activity following the oral administration of 600 mg of Compound I with and without food.

[0064] Administration, dosing, and sampling schedules were commensurate with those described in Examples 1 and 2. However, patients were administered 600 mg (rather than 900 mg) of Compound I with and without food. Patients were administered two tablets of 300 mg each. The tablets were prepared as described in Example 1.

[0065] Relative CETP activities (calculated as a percentage of baseline CETP activity) and standard deviations (SD) were measured, and the resulting data is summarized in Table 3.

Table 3 – Relative CETP Activity

Treatment Protocol	CETP Activity Relative to Pre-dose (standard deviation)							
	Pre-dose	1 h	2 h	4 h	6 h	8 h	24 h	Post-study
Fed	100 (0.0)	100.4 (1.9)	87.8 (9.2)	52.6 (13.4)	37.6 (6.6)	39.1 (8.3)	65.5 (5.7)	102.9 (7.3)
Fasted	100 (0.0)	102.1 (3.6)	99.6 (2.1)	96.5 (3.1)	89.5 (2.0)	87.8 (4.6)	92.6 (3.0)	100.4 (2.8)

[0066] A clear difference in CETP relative activity was observed when Compound I was administered with and without food, consistent with the results discussed in Example 2. Inhibition of CETP activity was much more marked in the fed treatment protocol as compared with the fasted treatment protocol. For example, between 4 and 24 hours post-dose, there was a significant decrease in CETP activity in the fed versus the fasted state. Specifically, the inhibition of CETP activity following the administration of Compound I with food reached its peak at 6 hours post-administration with 37.6% CETP activity relative to baseline. In contrast, the inhibition of CETP activity following the administration of Compound I without food reached its peak at 8 hours post-administration with 87.8% CETP activity relative to baseline. Such a decrease in the relative CETP activity following the administration of Compound I with food indicates increased bioavailability of the active form of the drug when administered with food as compared to the administration of the drug without food.

EXAMPLE 4

[0067] In a similar study to that described in Example 1, the effect of food on the absorption of the active form of Compound I in patients was identified in a study designed to compare the bioavailability of 600 mg of Compound I orally administered to Japanese male volunteers with and without food.

[0068] Administration, dosing, and sampling schedules were commensurate with those described in Example 1. However, patients were administered 600 mg (rather than 900 mg) of *S*-[2-([[(1-(2-ethylbutyl)cyclohexyl]carbonyl]amino)phenyl] 2-methylpropanethioate with and without food. Patients were administered two tablets of 300 mg each. The tablets were prepared as described in Example 1.

[0069] As observed in Example 1, pharmacokinetics parameters, such as maximum observed plasma concentration (C_{max}) and area under the plasma concentration-time curve from time zero to infinity ($AUC_{0-\infty}$), were affected by administration of Compound I with food. The C_{max} (mean) value after administration of 600 mg of Compound I was 1.029 $\mu\text{g}/\text{mL}$ when administered with food and only 0.316 $\mu\text{g}/\text{mL}$ when administered without food. The $AUC_{0-\infty}$ (mean) value after administration of 600 mg of Compound I was 10.458 $\mu\text{g h/mL}$ when administered with food and only 5.395 $\mu\text{g h/mL}$ when administered without food. Thus, the C_{max} and $AUC_{0-\infty}$ were about 3 and 2 times higher, respectively, when the patients were administered the drug with food as compared to without food.

[0070] The observed increases in the pharmacokinetic parameters when Compound I is administered with food indicate that the active form of the drug is more readily absorbed when administered with food, such as after a meal. Thus, the administration of Compound I with food results in an increase in the bioavailability of the active form of the drug when compared to the administration of the drug under fasted conditions.

[0071] All references, including publications, patent applications, and patents, cited herein are hereby incorporated by reference to the same extent as if each reference were individually and specifically indicated to be incorporated by reference and were set forth in its entirety herein.

[0072] The use of the terms “a” and “an” and “the” and similar referents in the context of describing the invention (especially in the context of the following claims) are to be construed to cover both the singular and the plural, unless otherwise indicated herein or clearly contradicted by context. The terms “comprising,” “having,” “including,” and “containing” are to be construed as open-ended terms (i.e., meaning “including, but not limited to,”) unless otherwise noted. Recitation of ranges of values herein are merely intended to serve as a shorthand method of referring individually to each separate value

falling within the range, unless otherwise indicated herein, and each separate value is incorporated into the specification as if it were individually recited herein. All methods described herein can be performed in any suitable order unless otherwise indicated herein or otherwise clearly contradicted by context. The use of any and all examples, or exemplary language (e.g., "such as") provided herein, is intended merely to better illuminate the invention and does not pose a limitation on the scope of the invention unless otherwise claimed. No language in the specification should be construed as indicating any non-claimed element as essential to the practice of the invention.

[0073] Preferred embodiments of this invention are described herein, including the best mode known to the inventors for carrying out the invention. Variations of those preferred embodiments may become apparent to those of ordinary skill in the art upon reading the foregoing description. The inventors expect skilled artisans to employ such variations as appropriate, and the inventors intend for the invention to be practiced otherwise than as specifically described herein. Accordingly, this invention includes all modifications and equivalents of the subject matter recited in the claims appended hereto as permitted by applicable law. Moreover, any combination of the above-described elements in all possible variations thereof is encompassed by the invention unless otherwise indicated herein or otherwise clearly contradicted by context.